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(71) Applicant: **SUNTORY LIMITED**
Kita-ku, Osaka-shi, Osaka 530 (JP)

(72) Inventors:
- **Adachi, Hideki**
  Asaka-shi, Saitama 351 (JP)
- **Tsujimoto, Masafumi**
  Asaka-shi, Saitama 351 (JP)
- **Arai, Hiroyuki**
  Arakawa-ku, Tokyo 116 (JP)
- **Inoue, Keizo**
  Kota-ku, Tokyo 135 (JP)

(74) Representative:
Wächtershäuser, Günter, Prof. Dr.
Patentanwalt,
Tal 29
80331 München (DE)

(54) Platelet activating factor acetylhdrolase, and gene thereof

(57) A protein having activities of a human platelet activating factor acetylhydrolase, and represented by an amino acid sequence represented by the following formula (I) or an amino acid sequence having homology therewith; and a DNA encoding the protein:

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Met Gly Val Asn Gln Ser Val Gly Phe Pro Pro Val Thr Gly Pro
His Leu Val Gly Cys Gly Asp Val Met Glu Gly Gln Asn Leu Gln
Gly Ser Phe Phe Arg Leu Phe Tyr Pro Cys Gln Lys Ala Glu Glu
Thr Met Glu Gln Pro Leu Trp Ile Pro Arg Tyr Glu Tyr Cys Thr
Gly Leu Ala Glu Tyr Leu Gln Phe Asn Lys Arg Cys Gly Gly Leu
Leu Phe Asn Leu Ala Val Gly Ser Cys Arg Leu Pro Val Ser Trp
Asn Gly Pro Phe Lys Thr Lys Asp Ser Gly Tyr Pro Leu Ile Ile
Phe Ser His Gly Leu Gly Ala Phe Arg Thr Leu Tyr Ser Ala Phe
Cys Met Glu Leu Ala Ser Arg Gly Phe Val Val Ala Val Pro Glu
His Arg Asp Arg Ser Ala Ala Thr Thr Tyr Phe Cys Lys Gln Ala
Pro Glu Glu Asn Gln Pro Thr Asn Glu Ser Leu Gln Glu Glu Trp
Ile Pro Phe Arg Arg Val Glu Glu Gly Glu Lys Glu Phe His Val
Arg Asn Pro Gln Val His Gln Arg Val Ser Glu Cys Leu Arg Val
Leu Lys Ile Leu Gln Glu Val Thr Ala Gly Gln Thr Val Phe Asn
Ile Leu Pro Gly Gly Leu Asp Leu Met Thr Leu Lys Gly Asn Ile
Asp Met Ser Arg Val Ala Val Met Gly His Ser Phe Gly Gly Ala
Thr Ala Ile Leu Ala Leu Ala Lys Glu Thr Gln Phe Arg Cys Ala
Val Ala Leu Asp Ala Trp Met Phe Pro Leu Glu Arg Asp Phe Tyr
Pro Lys Ala Arg Gly Pro Val Phe Phe Ile Asn Thr Glu Lys Phe

Gln Thr Met Glu Ser Val Asn Leu Met Lys Lys Ile Cys Ala Gln
His Glu Gln Ser Arg Ile Ile Thr Val Leu Gly Ser Val His Arg
Ser Gln Thr Asp Phe Ala Phe Val Thr Gly Asn Leu Ile Gly Lys
Phe Phe Ser Thr Glu Thr Arg Gly Ser Leu Asp Pro Tyr Glu Gly
Gln Glu Val Met Val Arg Ala Met Leu Ala Phe Leu Gln Lys His
Leu Asp Leu Lys Glu Asp Tyr Asn Gln Trp Asn Asn Leu Ile Glu
Gly Ile Gly Pro Ser Leu Thr Pro Gly Ala Pro His His Leu Ser
Ser Leu

(I)

Description

BACKGROUND OF THE INVENTION

a) Field of the Invention

This invention relates to a novel platelet activating factor acetylhydrolase, and a gene encoding the same.

b) Description of the Related Art

A platelet activating factor acetylhydrolase is an enzyme, which acts on a platelet activating factor (hereinafter abbreviated as "PAF") and eliminates its 2-acetyl group to deprive PAF of its activity. Since PAF is a mediator for inflammation which causes defluxion of tissue fluid through finer vessels, vasodilation, smooth muscle contraction, endothelial adhesion, activation of neutrophils, macrophages or eosinophilic leukocytes, or the like, PAF acetylhydrolase is usable as a preventive or therapeutic for various diseases caused by PAF.

Some reports have been made about PAF acetylhydrolase to date. For its use as a medicine, however, there is an outstanding desire for the provision of a PAF acetylhydrolase having higher purity and stronger action compared with conventional PAF acetylhydrolase. Further, from the viewpoint of safety, PAF acetylhydrolase derived form human being instead of an animal is desired.

SUMMARY OF THE INVENTION

With the foregoing in view, the present invention has as a primary object the provision of PAF acetylhydrolase which can fulfill the above-described desires.

Interested in the wide-spread distribution of PAF acetylhydrolase in animal organs such as the brain and kidneys, the present inventors chose the bovine liver as a source, and by various isolation and purification procedures, progressively increased the purity of PAF acetylhydrolase while placing a focus on its enzymatic activity. As a result, the present inventors have succeeded in obtaining bovine PAF acetylhydrolase as a pure product and further in determining its amino acid sequence. In addition, from the amino acid sequence of the PAF acetylhydrolase, a gene encoding the enzyme has been found by methods known *per* se in the art.

Moreover, using the bovine PAF acetylhydrolase cDNA, the present inventors have also succeeded in identifying the human PAF acetylhydrolase cDNA.

The present invention has been completed based on these findings, and provides a human PAF acetylhydrolase, which plays an important role as a PAF-inhibiting substance, and also a gene which encodes the enzyme and is important for the synthesis of the enzyme by genetic engineering.

The human PAF acetylhydrolase according to the present invention selectively degrades PAF and hence, is usable as medicines or biochemical reagents for the prevention and treatment of diseases caused by PAF, for example, diseases such as asthma, exudative tympanitis, hemorrhagic colitis and adult respiratory distress syndrome.

DETAILED DESCRIPTION OF THE INVENTION AND PREFERRED EMBODIMENTS

The human PAF acetylhydrolase according to the present invention can be prepared as will be described next. PAF acetylhydrolase is first collected from an animal. From the PAF acetylhydrolase, the animal PAF acetylhydrolase cDNA is determined. Using the animal PAF acetylhydrolase cDNA, the human PAF acetylhydrolase cDNA is detected from a human gene library. The human PAF acetylhydrolase cDNA is inserted in an appropriate vector and then cultured in an adequate host organism, whereby the human PAF acetylhydrolase is obtained.

Upon practice of the present invention, it is first necessary to obtain animal PAF acetylhydrolase from an organ of an animal such as the brain, liver or kidneys by purifying it through repetitions of known isolation and purification procedures while using PAF acetylhydrolase activity as an index. A description will hereinafter be made of a process for obtaining PAF acetylhydrolase by using a bovine liver as an example.

As the bovine liver to be used as a source, one obtained from a bovine immediately after its slaughter is preferred.

After the bovine liver is first washed with an appropriate buffer (for example, 10 mM Tris-HCl buffer containing 250 mM sucrose and 1 mM EDTA and having a pH of 7.4), it is homogenized with the same buffer. The homogenate is then centrifuged to obtain a soluble fraction.

Making combined use of hydrophobic chromatography, ion exchange chromatography, adsorption chromatography, gel filtration chromatography and the like, the soluble fraction is purified until a single peak is observed by Mono Q FPLC, so that PAF acetyl hydrolase can be obtained.

Incidentally, PAF acetylhydrolase activity which is used as an index for the selective collection of the PAF-acetyl-

hydrolase-containing fraction can be determined, for example, by the method disclosed in Japanese Patent Application Laid-Open (Kokai) No. HEI 7-39373.

With respect to the bovine PAF acetylhydrolase obtained in the above-described manner, its amino acid sequence was investigated by a method known *per se* in the art. As a result, the amino acid sequence has been found to be represented by the following formula (III):

Met Gly Val Asn Gln Ser Val Ser Phe Pro Pro Val Thr Gly Pro
His Leu Val Gly Cys Gly Asp Val Met Glu Gly Gln Ser Leu Gln
Gly Ser Phe Phe Arg Leu Phe Tyr Pro Cys Gln Glu Ala Glu Glu
Thr Ser Glu Gln Pro Leu Trp Ile Pro Arg Tyr Glu Tyr Cys Ala
Gly Leu Ala Glu Tyr Leu Lys Phe Asn Lys Arg Trp Gly Gly Leu
Leu Phe Asn Leu Gly Val Gly Ser Cys Arg Leu Pro Val Ser Trp
Asn Gly Pro Phe Lys Thr Lys Asp Ser Gly Tyr Pro Leu Ile Ile
Phe Ser His Gly Met Gly Ala Phe Arg Thr Val Tyr Ser Ala Phe
Cys Met Glu Leu Ala Ser Arg Gly Phe Val Val Ala Val Pro Glu
His Arg Asp Gly Ser Ala Ala Ala Thr Cys Phe Cys Lys Gln Thr
Pro Glu Glu Asn Gln Pro Asp Asn Glu Ala Leu Lys Glu Glu Trp
Ile Pro His Arg Gln Ile Glu Glu Gly Glu Lys Glu Phe Tyr Val
Arg Asn Tyr Gln Val His Gln Arg Val Ser Glu Cys Val Arg Val
Leu Lys Ile Leu Gln Glu Val Thr Ala Gly Gln Ala Val Leu Asn
Ile Leu Pro Gly Gly Leu Asp Leu Met Thr Leu Lys Gly Gly Ile
Asp Val Ser Arg Val Ala Val Met Gly His Ser Phe Gly Gly Ala
Thr Ala Ile Leu Ala Leu Ala Lys Glu Met Gln Phe Arg Cys Ala

Val Ala Leu Asp Ala Trp Met Phe Pro Leu Glu His Asp Phe Tyr
Pro Thr Ala Arg Gly Pro Ile Phe Phe Ile Asn Ala Glu Lys Phe
Gln Thr Val Glu Thr Val Asn Leu Met Lys Lys Ile Cys Asp Gln
His His Gln Ser Arg Ile Ile Thr Val Leu Gly Ser Val His Arg
Ser Leu Thr Asp Phe Val Phe Val Ala Gly Asn Trp Ile Ser Lys
Phe Phe Ser Ser His Thr Arg Gly Ser Leu Asp Pro Tyr Glu Gly
Gln Glu Thr Val Val Arg Ala Met Leu Ala Phe Leu Gln Lys His
Leu Asp Leu Lys Glu Asp Tyr Asp Gln Trp Asn Asn Phe Ile Glu
Gly Ile Gly Pro Ser Leu Thr Pro Gly Ala Pro His His Leu Ser
Ser Leu

(III)

Further, from the peptide sequence of the bovine PAF acetylhydrolase of the formula (III), a gene encoding the enzyme was determined by a method known *per se* in the art. The gene (hereinafter called the "bovine PAF acetylhydrolase cDNA") has been found to be identified by the following formula (IV):

GTCGACCCACGCGTCCGAGTTGACCGT
CTGGGCTGTTTCTGAGGGTCAACGTGACTCGCCGTCAAGTTCAGCCACTGCCCAAGTCGT
CGTTCAGTTCAGTTGGTTATGAG ATG GGG GTC AAC CAG TCT GTG AGC TTC
CCA CCC GTC ACG GGA CCC CAC CTC GTA GGC TGT GGG GAT GTG ATG
GAG GGT CAG AGC CTC CAG GGC AGC TTC TTT CGA CTG TTC TAC CCG
TGC CAA GAG GCA GAG GAG ACC TCG GAG CAG CCC CTG TGG ATT CCC
CGC TAT GAG TAC TGC GCT GGC CTG GCC GAA TAC CTA AAG TTT AAT
AAG CGC TGG GGG GGG TTA CTG TTC AAC CTG GGT GTG GGA TCT TGT
CGC CTG CCT GTT AGC TGG AAT GGC CCC TTT AAA ACA AAG GAC TCT
GGA TAC CCC TTG ATC ATC TTC TCT CAT GGC ATG GGA GCC TTC AGG

ACA GTG TAT TCA GCC TTC TGC ATG GAG CTG GCT TCT CGT GGC TTT

GTG GTT GCT GTA CCA GAG CAC AGG GAT GGG TCA GCT GCG GCC ACC

TGT TTC TGC AAG CAG ACC CCA GAG GAG AAC CAG CCT GAC AAT GAG

GCC CTG AAG GAG GAA TGG ATC CCC CAC CGT CAA ATT GAG GAA GGG

GAG AAG GAA TTC TAT GTT CGG AAC TAC CAG GTG CAT CAG AGG GTG

AGC GAG TGT GTG AGG GTG TTG AAG ATC CTA CAA GAG GTC ACT GCT

GGG CAG GCC GTT CTC AAC ATC TTG CCT GGC GGA TTG GAT CTG ATG

ACC TTG AAG GGC GGC ATT GAC GTG AGC CGT GTG GCT GTA ATG GGA

CAT TCA TTT GGA GGG GCC ACA GCT ATT CTG GCC TTG GCC AAG GAG

ATG CAA TTT AGG TGT GCT GTG GCT TTG GAC GCT TGG ATG TTT CCT

CTG GAG CAT GAC TTT TAC CCC ACG GCC CGA GGC CCT ATC TTC TTT

ATC AAT GCT GAG AAG TTC CAG ACA GTG GAG ACT GTC AAC TTG ATG

AAA AAG ATT TGT GAC CAG CAC CAC CAA TCC AGG ATC ATA ACT GTC

CTT GGT TCT GTT CAT CGG AGT CTA ACC GAC TTT GTT TTT GTG GCT

GGT AAC TGG ATT AGT AAA TTC TTC TCC AGT CAC ACC CGT GGA AGC

TTG GAC CCC TAT GAA GGT CAG GAG ACC GTG GTG CGG GCC ATG TTG

GCC TTC CTG CAG AAG CAT CTT GAC CTG AAA GAG GAC TAT GAC CAG

TGG AAC AAC TTC ATT GAA GGC ATT GGC CCA TCA CTG ACC CCA GGG

GCC CCA CAC CAT CTG TCC AGC CTG TAG GCACAACTGGTCATCTTGTGGAAG

GTCCCTGAGCTGAGTTCCCGTGTGGGGCCTGCCCAGGGATACCCTTGGCCTCCTATCAGG

AAGTGATTGCCATGACCCTTCTGTGTTGATTGAGAGGATATAATCACACTGCTGATTGGT

AACGGGGTACTTGGATTCTCAGACTTGTCGATCTTAAACTCATGTTGGGACTTGGGTTCA

CTTACTGATGGGCAAACGGGCATTCTGAGGACTGAGCCTTAATGGTATGGAGAACAAACA

GTGGGATGGGGCTGGGGAAGATCTAAGCCCTAAGCTGGGCACTATGAGCCCTATAAACCC

AACCAGCCAACACCCTCACCTTGGGCAAGTATGACTTCTGCAGGTCGACTCT

(IV)

To obtain human PAF acetylhydrolase from the bovine PAF acetylhydrolase cDNA obtained as described above, the human gene library is screened by a method known *per se* in the art while using the bovine PAF acetylhydrolase cDNA as a template.

Described specifically, the bovine PAF acetylhydrolase cDNA is labeled, for example, by incorporating fluorescein-

12-dUTP through PCR. By the colony hybridization technique that selects each positive colony by ECL (Enhanced Chemiluminescence; Amersham K.K.), colonies containing the human PAF acetylhydrolase cDNA can be obtained.

The human PAF acetylhydrolase cDNA obtained as described above has been found to be identified by the following formula (II):

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                                                GCAGGTCTCGACCCACGCGTCCGCGGACGCGTGGG

          CGAGAAGTGCTTCCAAGCGTCCATTTTGAGCCTTGGAAACTACGACGACCAAAGGGCCAC

          GGGTTCCTGGGTCGTTTCTCATTTCCGTCGAGTTAAACGTCTGGGGCTGCTTCTGAGGAA

          TCAGCTTGGCTGGCCAGCAAGTTCAGCTCCGGCAAGTCATTTGATTCACCCGGTGATGAA

          ATG GGG GTC AAC CAG TCT GTG GGC TTT CCA CCT GTC ACA GGA CCC

          CAC CTC GTA GGC TGT GGG GAT GTG ATG GAG GGT CAG AAT CTC CAG

          GGG AGC TTC TTT CGA CTC TTC TAC CCC TGC CAA AAG GCA GAG GAG

          ACC ATG GAG CAG CCC CTG TGG ATT CCC CGC TAT GAG TAC TGC ACT

          GGC CTG GCC GAG TAC CTG CAG TTT AAT AAG CGC TGC GGG GGC TTG

          CTG TTC AAC CTG GCG GTG GGA TCT TGT CGC CTG CCT GTT AGC TGG

          AAT GGC CCC TTT AAG ACA AAG GAC TCT GGA TAC CCC TTG ATC ATC
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TTC TCC CAT GGC CTA GGA GCC TTC AGG ACT TTG TAT TCA GCC TTC

TGC ATG GAG CTG GCC TCA CGT GGC TTT GTG GTT GCT GTG CCA GAG

CAC AGG GAC CGG TCA GCG GCA ACC ACC TAT TTC TGC AAG CAG GCC

CCA GAA GAG AAC CAG CCC ACC AAT GAA TCG CTG CAG GAG GAA TGG

ATC CCT TTC CGT CGA GTT GAG GAA GGG GAG AAG GAA TTT CAT GTT

CGG AAT CCC CAG GTG CAT CAG CGG GTA AGC GAG TGT TTA CGG GTG

TTG AAG ATC CTG CAA GAG GTC ACT GCT GGG CAG ACT GTC TTC AAC

ATC TTG CCT GGT GGC TTG GAT CTG ATG ACT TTG AAG GGC AAC ATT

GAC ATG AGC CGT GTG GCT GTG ATG GGA CAT TCA TTT GGA GGG GCC

ACA GCT ATT CTG GCT TTG GCC AAG GAG ACC CAA TTT CGG TGT GCG

GTG GCT CTG GAT GCT TGG ATG TTT CCT CTG GAA CGT GAC TTT TAC

CCC AAG GCC CGA GGA CCT GTG TTC TTT ATC AAT ACT GAG AAA TTC

CAG ACA ATG GAG AGT GTC AAT TTG ATG AAG AAG ATA TGT GCC CAG

CAT GAA CAG TCT AGG ATC ATA ACC GTT CTT GGT TCT GTT CAT CGG

AGT CAA ACT GAC TTT GCT TTT GTG ACT GGC AAC TTG ATT GGT AAA

TTC TTC TCC ACT GAA ACC CGT GGG AGC CTG GAC CCC TAT GAA GGG

CAG GAG GTT ATG GTA CGG GCC ATG TTG GCC TTC CTG CAG AAG CAC

CTC GAC CTG AAA GAA GAC TAT AAT CAA TGG AAC AAC CTT ATT GAA

GGC ATT GGA CCG TCG CTC ACC CCA GGG GCC CCC CAC CAT CTG TCC

AGC CTG TAG GCACAACTGGCCATTTGTAAAGTCACTTCAGCCAAGTTTTCATTTGGG

AGCTACCCAAGGGCACCCATGAGCTCCTATCAAGAAGTGATCAACGTGACCCCTTTTCAC

AGATTGAAAGGTGTAATCACACTGCTGCTTGGATAACTGGGTACTTTGATCTTAGATTTG

ATCTTAAAATCACTTTGGGACTGGGATCCCTTGCTGATTGACAAACAGACTTTCTGGGAC

CTTGATGGAGTGGGGAACAAGCAGTAGAGTGGGACTGGGGGAGACCCAGGCCCCGGGCTG

AGCACTGTGAGGCCTGGATGTGAAGACTCAGCCCAGCGAAGCTCATTCCCTTACCCCCGG

CCAGTGCTGCTGCTTCAGTGGAAGAGATGAAGCCAAAGGACAGAATGAAAATCCCTACCT

TCAGAGACTCTAGCCCAGCCCAACACCATCTCTTCCTACCTCTCAGCCTTCTCCCTCCCC

AGGGCCACTTGTTGAAGTCTGAGCACTTTATGTAAATTTCTAGGTGTGAGCCGTGATCAC

ATTTTCTATTTATTTCCAAGTCTTCTCATTGTATGGAACATAGTACTACTTATACTTACA

GTAGTAAGTTATACTTGTGAGCCCACAGAGTGGCAGACAGCATGGCTCTCACAGCACAGG

GAGAAAAACTGAGGTACACAGAGGTACCTCAGAAGCTCTGGATGTCTTTGGGGGTTTTGC

TAAGTGTATCTTGATAGGAAACAACAAAAGCAGGTTGAGATGGGGAAGATGACAGAACAA

CAGTGTTAAATGGCCATTTGCACAGGCCTTTGCCACAACAGAGAAGTAGTTTGGTCAGCT

AAAACTCAGCTGCAGCCTGGACAGTAGAGCGAGACCCCATCTTAAAAATAAAGAAGGCTG

GGCGTGGTGGCTCATGCCTGTAATCCCAGCACTTTGGGAGGCCAAGGCAGGCAGATCACT

TAAGGCCAGGAGTTCAAGACCACCTGGCCAACATGGTGAAACCCCGTCTCTACTAAAAAT

ACAAAAAATTAGCCTGGCGTAATGGCAGGCGCCTATAATCCCAGCTACTCAGGAGGCTGA

AGCAGAAGAATCACTTGAACCTAGGAGGCGGAGGTTGCAGTGAGTCAAGATCGCGCCACT

GCACTCCAGCCTGGGTGACAGAGCAAGACTCTGTCTT

(II)

Following conventional procedures, the human PAF acetylhydrolase cDNA obtained as described above is next introduced in an appropriate vector plasmid, and host cells such as mammal cells are then transformed by a commonly-employed recombinant DNA technique to express the human PAF acetylhydrolase. The expression of the human PAF acetylhydrolase can be confirmed by a western blot technique which makes use of an anti-human PAF acetylhydrolase antibody. The introduction into the plasmid, the establishment of the transformed strain, the culture of the strain and the like can be conducted by the general recombinant DNA technology.

From expression systems known to artisans, a suitable expression system can be selected for use in the present invention. It is possible to improve the efficiency of secretion and the level of expression by adding or improving a signal sequence and/or choosing an appropriate host. Although no particular limitation is imposed on host cells, illustrative examples include cultured cells of bacteria, yeasts, other fungi, human and other animals, and cultured cells of plants. Namely, the polynucleotide according to the present invention is inserted in a suitable expression vector, for example, pUC-PL-cI vector, the expression vector is introduced in adequate host cells, for example, *E. Coli* W3110 or the like, and the host cells are then cultured. The target human PAF acetylhydrolase can thereafter be collected as a protein from the thus-obtained cultured matter (cells or culture medium).

As the host, a procaryote or an eucaryote can be used. Usable examples of the procaryote include bacteria, especially *Escherichia coli* and *Bacillus* bacteria, for example, *B. subtilis.* On the other hand, usable examples of the eucaryote include eucaryotic microorganisms such as yeasts, for example, *Saccharomyces* yeasts, especially *s. Servisiae;* insect cells such as armyworm *(Spodoptera Frugiperda)* cells and silkworm *(Bombyx mori)* cells; and animal cells such as human cells, monkey cells and mouse cells, especially monkey cells, for example, COS1 and COS 7.

Usable examples of the expression vector include plasmids, pharges, phargemids, viruses [baculoviruses (for insect cells), vaccinia viruses (for animal cells)]. The promoter in the expression vector is selected depending on the host cells. For examples, lac promoters, trp promoters, trc promoters and the like can be used as promoters for bacteria; and adh 1 promoters, pgk promoters and the like can be used as promoters for yeasts. Further, baculovirus polyhedrin promoters can be mentioned as promoters for insects; and early and late promoters of *Simian virus* 40 (SV40) can be mentioned as promoters for animal cells.

When an enhancer is used, for example, the enhancer of SV40 is inserted either upstream or downstream of the gene.

The transformation of the host by the expression vector can be conducted by a common method known *per se* in the art. Such methods are disclosed, for example, in "Current Protocols in Molecular Biology", John Wiley & Sons, Inc.

The culture of the transformants can also be conducted by a usual method. The purification of the human PAF acetylhydrolase from the cultured matter can be conducted following procedures commonly employed for the isolation and purification of proteins, for example, by ultrafiltration and/or one or more of various column chromatographic procedures, for example, chromatography making use of "Sepharose".

In the above-described manner, the human PAF acetylhydrolase can be advantageously obtained. The human PAF acetylhydrolase according to the present invention is represented by the following formula (I):

Met Gly Val Asn Gln Ser Val Gly Phe Pro Pro Val Thr Gly Pro
His Leu Val Gly Cys Gly Asp Val Met Glu Gly Gln Asn Leu Gln
Gly Ser Phe Phe Arg Leu Phe Tyr Pro Cys Gln Lys Ala Glu Glu
Thr Met Glu Gln Pro Leu Trp Ile Pro Arg Tyr Glu Tyr Cys Thr
Gly Leu Ala Glu Tyr Leu Gln Phe Asn Lys Arg Cys Gly Gly Leu
Leu Phe Asn Leu Ala Val Gly Ser Cys Arg Leu Pro Val Ser Trp
Asn Gly Pro Phe Lys Thr Lys Asp Ser Gly Tyr Pro Leu Ile Ile
Phe Ser His Gly Leu Gly Ala Phe Arg Thr Leu Tyr Ser Ala Phe
Cys Met Glu Leu Ala Ser Arg Gly Phe Val Val Ala Val Pro Glu
His Arg Asp Arg Ser Ala Ala Thr Thr Tyr Phe Cys Lys Gln Ala
Pro Glu Glu Asn Gln Pro Thr Asn Glu Ser Leu Gln Glu Glu Trp
Ile Pro Phe Arg Arg Val Glu Glu Gly Glu Lys Glu Phe His Val
Arg Asn Pro Gln Val His Gln Arg Val Ser Glu Cys Leu Arg Val
Leu Lys Ile Leu Gln Glu Val Thr Ala Gly Gln Thr Val Phe Asn

Ile Leu Pro Gly Gly Leu Asp Leu Met Thr Leu Lys Gly Asn Ile
Asp Met Ser Arg Val Ala Val Met Gly His Ser Phe Gly Gly Ala
Thr Ala Ile Leu Ala Leu Ala Lys Glu Thr Gln Phe Arg Cys Ala
Val Ala Leu Asp Ala Trp Met Phe Pro Leu Glu Arg Asp Phe Tyr
Pro Lys Ala Arg Gly Pro Val Phe Phe Ile Asn Thr Glu Lys Phe
Gln Thr Met Glu Ser Val Asn Leu Met Lys Lys Ile Cys Ala Gln
His Glu Gln Ser Arg Ile Ile Thr Val Leu Gly Ser Val His Arg
Ser Gln Thr Asp Phe Ala Phe Val Thr Gly Asn Leu Ile Gly Lys
Phe Phe Ser Thr Glu Thr Arg Gly Ser Leu Asp Pro Tyr Glu Gly
Gln Glu Val Met Val Arg Ala Met Leu Ala Phe Leu Gln Lys His
Leu Asp Leu Lys Glu Asp Tyr Asn Gln Trp Asn Asn Leu Ile Glu
Gly Ile Gly Pro Ser Leu Thr Pro Gly Ala Pro His His Leu Ser
Ser Leu

(I)

The human PAF acetylhydrolase selectively degrades PAF and oxidized phospholipids and has physiologically active effects such anti-inflammatory effects.

Needless to say, the human PAF acetylhydrolase according to the present invention is not limited to the peptide of the formula (I) but includes peptides having homology therewith, namely, peptides having the same function as the peptide represented by the formula (I) despite substitution, deletion, addition or the like of amino acids at parts of their sequences.

The bovine PAF acetylhydrolase represented by the formula (III) may be contemplated to be available by gene manipulation in a similar manner as the human PAF acetylhydrolase. As a matter of fact, however, the bovine PAF acetylhydrolase cannot be obtained unless eucaryotic host cells are used.

To obtain the bovine PAF acetylhydrolase by gene manipulation, it is therefore necessary to employ as host cells those derived from an eucaryote and to select and use a vector compatible with the host cells.

An antibody against the human PAF acetylhydrolase or bovine PAF acetylhydrolase (which may hereinafter be collectively called the "PAF acetylhydrolase") according to the present invention can also be obtained following usual procedures.

Described specifically, the antibody can be obtained by sensitizing an animal such as a rabbit with the PAF acetylhydrolase, separating its serum and, if necessary, purifying an immunoglobulin fraction from the serum. To enhance the sensitizing ability of the enzyme in this case, the enzyme in a form bound on a carrier protein such as bovine serum albumin (BSA) or methyl BSA may be used as an immunogen.

Upon sensitizing an animal, the enzyme can also be used together with Freund's complete adjuvant (FCA) or Freund's incomplete adjuvant (FICA) to increase the production of the antibody. It is desired to conduct the sensitization of the animal twice or more. The frequency of sensitization can be determined while checking the antibody titer of the serum by test sampling of blood. The whole blood of an immune animal may be used by slaughtering it as needed. As an alternative, an immune animal may be subjected to booster sensitization as many times as needed to maintain a constant antibody titer, and blood samples may be collected in small quantities as needed for immediate use. It is also possible to obtain a monoclonal antibody in a usual manner by sensitizing a mouse with the enzyme and then forming hybridomas from spleen cells and myeloma cells of the sensitized mouse.

The present invention will hereinafter be described in further detail by the following examples and reference examples. It is however to be noted that the present invention are by no means limited by or to these examples.

Referential Example 1

Measurement of PAF Acetylhydrolase Activity

(1) Using unlabeled lyso PAF (product of Bachem Feinchemikalien AG), 1-O-[1-^{14}C]hexadecyl-lyso PAF (product of New England Nuclear Company; hereinafter called the "labeled lyso PAF") was diluted to 4,000 dpm/nmol.

On the other hand, 1-O-hexadecyl-2-[^{3}H-acetyl]-sn-glycero-3-phosphocholine (hereinafter called "^{3}H-acetyl PAF") was diluted to 3,200 dpm/nmol with the unlabeled lyso PAF.

A standard culture system for the measurement of PAF acetylhydrolase was composed of 50 mM Tris-HCl (pH 7.4), 5 mM EDTA, 5 mM 2-mercaptoethanol (2-ME) and 20 nmol ^{3}H-acetyl PAF. The total volume of the sample was 0.25 mℓ.

(2) Measurement of PAF acetylhydrolase activity was conducted by culturing a test sample in the above-described standard culture system at 37°C for 30 minutes, adding 2.5 me of chloroform/methanol (4:1 V/V) and 0.25 mℓ of water to terminate the reaction, and then measuring the radioactivity of a small amount (0.6 mℓ) of each upper layer to determine the amount of the acetate liberated from the ^{3}H-acetyl PAF.

Example 1

Obtainment of Bovine PAF Acetylhydrolase

(1) A fresh bovine liver was purchased from a slaughterhouse and was then treated within 3 hours of the slaughter. Treatments were all conducted at 0 to 4°C. The liver was homogenized in a Waring blender subsequent to the addition of a homogenizing buffer [10 mM Tris-HCl (pH 7.4), 250 mM sucrose, 1 mM EDTA] in an amount 5 times as much as the liver. The resulting homogenate was centrifuged for 30 minutes under 100,000 x g, followed by the removal of a solid portion. The resultant supernatant was centrifuged further for 1 hour under 100,000 x g, whereby a dissolved portion was obtained (supernatant portion)

(2) The supernatant portion obtained through the procedures (1) was adjusted to 1 M with NaCl. Subsequent to stirring for 15 minutes, the solution was loaded on a "BUTYL TOYOPEARL 650 M" column which had been equilibrated beforehand with a buffer composed of 50 mM Tris-HCl (pH 7.4), 1 mM EDTA and 1 M NaCl. After the column was washed with the same buffer, proteins were eluted with a linear gradient of NaCl (1 to 0 M). PAF acetylhydrolase activity was eluted as a single peak in 1 to 0 M NaCl fractions.

(3) Active fractions from the "BUTYL TOYOPEARL" column were loaded on a "Q-Sepharose" column which had been equilibrated with 10 mM Tris-HCl (pH 7.4), 1 mM EDTA and 20% (V/V) glycerol (buffer A). The column was washed with the buffer A. Proteins were eluted with a linear gradient of NaCl (0 to 500 mM) in the buffer A. The activity was observed in a fraction eluted with about 300 mM NaCl.

(4) The active fraction from the "Q-Sepharose" column was concentrated to about 6 mℓ in an "Amicon ultrafiltration cell" in which "YM-10" membranes were used. The thus-concentrated fraction was loaded on a "Biogel A-1.5 m" gel filtration column which had been equilibrated beforehand with 10 mM Tris-HCl (pH 7.4), 200 mM NaCl, 5 mM 2-ME, 20% (V/V) glycerol and 0.5 % (W/V) "CHAPS" (buffer B). The activity was eluted as a single peak in a fraction corresponding to a molecular weight of about 40 kDa.

(5) The active fraction from the "Biogel-A 1.5 m" column was loaded on a hydroxyapatite column which had been equilibrated beforehand with 10 mM Tris-HCl (pH 7.4), 5 mM 2-ME, 20% (V/V) glycerol and 0.5% (W/V) "CHAPS" (buffer C). Proteins were eluted with a linear gradient which ranged from the buffer C alone to a buffer C containing 150 mM KH_2PO_4. The activity was observed in a fraction which was eluted with about 50 mM KH_2PO_4.

(6) The active fraction from the hydroxyapatite column was dialyzed against the buffer C, and was then loaded on an "FPLC Mono Q HR 5/5" column which had been equilibrated beforehand with the buffer C. Proteins were eluted by a linear gradient of NaCl (0 to 500 mM) in the buffer C. The activity was observed in a fraction which was eluted with 250 mM NaCl, and a protein in the fraction was obtained as purified bovine PAF acetylhydrolase.

The total proteins, total activities, purification degrees (in terms of times) and the like in the individual purification steps described above are tabulated below:

Step	Total proteins (mg)	Total activity (μmol/min)	Activity per weight (nmon/min/mg)	Degree of purification (times)	Yield (%)
Cytoplasm	46000	73.5	1.6	1	100
BUTYL TOYOPEAL	680	16.3	24	15	22
Q Sepharose FF	72.4	8.96	124	78	12
Biogel A-1.5 m	6.93	7.38	1060	670	10
Hydroxyapatite	3.45	5.29	1530	960	7.2
Mono Q FPLC	0.3	2.16	7200	4500	2.9

Example 2

Determination of Amino Acid Sequence of Bovine PAF Acetylhydrolase

(1) About 0.2 mg of the purified PAF acetylhydrolase obtained in Example 1 was reduced with 1 mg of dithiothreitol at room temperature for 2 hours, followed by the S-alkylation with 0.6% (W/V) 4-vinylpyridine at room temperature for 2 hours.

Using a 4.6 mm x 250 mm "Vydak 304-1251 C_4" column which had been equilibrated beforehand with 20% (V/V) acetonitrile containing 0.1% (V/V) trifluoroacetic acid, the reaction mixture was subjected to reverse phase high-performance liquid chromatography (HPLC). Proteins were then eluted with a linear gradient of acetonitrile (20 to 85% V/V) which contained 0.1% (V/V) trifluoroacetic acid.

(2) 40 kDa polypeptide, which had been purified by the HPLC, was dialyzed against a lysylendopeptidase digestive buffer [0.5 M Tris-HCl (pH 8.5) and 4 M urea]. Next, 1 μg of a lysylendopeptidase was added to the sample. After the reaction mixture was incubated for 18 hours at 37°C, the reaction mixture was fractionated by reverse phase HPLC through a 4.6 mm x 250 mm "Vydak 304-1251 C_4" column while using a linear gradient of acetonitrile (5 to 70% V/V) which contained 0.1% (V/V) trifluoroacetic acid.

(3) The amino acid sequence of a peptide fragment obtained by the reverse phase HPLC was determined by an automated sequencer ("Model 477A", trade name; manufactured by Applied Biosystems, Inc.).

The base sequence of the bovine PAF acetylhydrolase, which was determined from the amino acid sequence of the peptide fragment, was as shown above by the formula (III).

Further, from the peptide sequence (III) of the bovine PAF acetylhydrolase, a gene encoding the enzyme was determined by a method known *per se* in the art. The gene was found to be represented by the formula (IV).

Example 3

Cloning of Non-active Human PAF Acetylhydrolase cDNA

Using as a template the bovine PAF acetylhydrolase cDNA obtained in Example 2, fluorescein-12-dUTP was incorporated in 500,000 clones of each of a fetal human liver cDNA library (pRc/CMV) and a human brain cDNA library (pCMV SPORTS) by PCR. The clones were then subjected to colony hybridization while detecting the labeling reagent by ECL, whereby cloning was conducted. As a result, a single positive clone was obtained from the human brain library.

A plasmid DNA was prepared and the base sequence was determined. The clone was a full-length clone which contained ATG encoding initiating methionine. Encoding 43 N-terminal amino acids were the same as the corresponding amino acids in the sequence of the bovine PAF acetylhydrolase up to the 40th amino acid, and there was poly A at the 3' end. A more accurate determination of the base sequence was conducted. As a result, the cDNA was found to consist of 2188 bp and to contain an ORF (open reading frame) consisting of 253 amino acids. Compared with the bovine PAF acetylhydrolase cDNA, 140 amino acids had been deleted. The segment of the deleted 140 amino acids contains a "catalytic triad" of serine, histidine and aspartic acid, which exhibits catalytic activity. The cDNA is therefore not believed to have PAD acetylhydrolase activity.

Hence, a primer was synthesized at positions flanking the deleted region, and PCR was conducted using the library DNA as a template. From the human brain cDNA, two bands were obtained, one corresponding to the above-described cDNA with the 140 amino acids deleted, and the other to a cDNA having substantially the same length as the bovine PAF acetylhydrolase cDNA. From the foregoing, the human brain library DNA was expected to contain, in addition to the above-obtained cDNA, a human PAF acetylhydrolase cDNA which is actually equipped with PAF acetylhydrolase activity.

Example 4

Cloning of Human PAF Acetylhydrolase cDNA

The human brain cDNA library was diluted to give 2000 clones per well, followed by incubation on 5 96-well plates. Subpools consisting of 10 wells were prepared, and positive pools were determined by PCR (Pool Nos. 10, 20, 28, and 38). With respect to these subpools, PCR was conducted well after well, so that positive pools were confirmed (Pool Nos. 10-5, 20-10, and 38-12).

Concerning these pools, incubation was conducted on plates subsequent to dilution. Using the non-active human PAF acetylhydrolase cDNA as a probe, cloning was attempted by hybridization. Labeling of the DNA was conducted with fluorescein 12-dUTP by PCR, and detection was carried out by ECL. Positive colonies were obtained from Pool

Nos. 10-5 and 20-10. Plasmid DNAs of these clones were replicated, and their base sequences were then determined. As a result, a human PAF acetylhydrolase cDNA represented by the formula (II) was obtained from the clones of Pool Nos. 10-5.

Based on the resultant cDNA, the amino acid sequence of the human PAF acetylhydrolase was determined. It was found to be represented by the formula (I). Up to 88%, the sequence was the same as that of the bovine PAF acetylhydrolase (346/392 amino acids). On the other hand, it was 42% identical to that of the plasma human PAF acetylhydrolase (162/392 amino acids).

Further, the above cDNA was incorporated in the pUC-PI-cI vector, introduced in *E. coli* W3110 and then subjected to expression. A band, which corresponded to a protein having a molecular weight of 42 kDa, was detected by SDS-PAGE.

The protein was investigated for activity. Human PAF acetylhydrolase activity was confirmed.

[Sequence Listing]

SEQ. ID. No.: 1

SEQ. LENGTH: 392

SEQ. TYPE: amino acid

MOLECULE TYPE: peptide

ORIGINAL SOURCE:

ORGANISM: bovine (*Bos taurus*)

SEQUENCE DESCRIPTION:

Met	Gly	Val	Asn	Gln	Ser	Val	Ser	Phe	Pro	Pro	Val	Thr	Gly	Pro
1				5				10						15
His	Leu	Val	Gly	Cys	Gly	Asp	Val	Met	Glu	Gly	Gln	Ser	Leu	Gln
				20				25						30
Gly	Ser	Phe	Phe	Arg	Leu	Phe	Tyr	Pro	Cys	Gln	Glu	Ala	Glu	Glu
				35				40						45
Thr	Ser	Glu	Gln	Pro	Leu	Trp	Ile	Pro	Arg	Tyr	Glu	Tyr	Cys	Ala
				50				55						60
Gly	Leu	Ala	Glu	Tyr	Leu	Lys	Phe	Asn	Lys	Arg	Trp	Gly	Gly	Leu
				65				70						75
Leu	Phe	Asn	Leu	Gly	Val	Gly	Ser	Cys	Arg	Leu	Pro	Val	Ser	Trp
				80				85						90
Asn	Gly	Pro	Phe	Lys	Thr	Lys	Asp	Ser	Gly	Tyr	Pro	Leu	Ile	Ile
				95				100						105
Phe	Ser	His	Gly	Met	Gly	Ala	Phe	Arg	Thr	Val	Tyr	Ser	Ala	Phe
				110				115						120

Cys Met Glu Leu Ala Ser Arg Gly Phe Val Val Ala Val Pro Glu
125 130 135
His Arg Asp Gly Ser Ala Ala Ala Thr Cys Phe Cys Lys Gln Thr
140 145 150

Pro Glu Glu Asn Gln Pro Asp Asn Glu Ala Leu Lys Glu Glu Trp
155 160 165
Ile Pro His Arg Gln Ile Glu Glu Gly Glu Lys Glu Phe Tyr Val
170 175 180
Arg Asn Tyr Gln Val His Gln Arg Val Ser Glu Cys Val Arg Val
185 190 195
Leu Lys Ile Leu Gln Glu Val Thr Ala Gly Gln Ala Val Leu Asn
200 205 210
Ile Leu Pro Gly Gly Leu Asp Leu Met Thr Leu Lys Gly Gly Ile
215 220 225
Asp Val Ser Arg Val Ala Val Met Gly His Ser Phe Gly Gly Ala
230 235 240
Thr Ala Ile Leu Ala Leu Ala Lys Glu Met Gln Phe Arg Cys Ala
245 250 255
Val Ala Leu Asp Ala Trp Met Phe Pro Leu Glu His Asp Phe Tyr
260 265 270
Pro Thr Ala Arg Gly Pro Ile Phe Phe Ile Asn Ala Glu Lys Phe
275 280 285
Gln Thr Val Glu Thr Val Asn Leu Met Lys Lys Ile Cys Asp Gln
290 295 300
His His Gln Ser Arg Ile Ile Thr Val Leu Gly Ser Val His Arg
305 310 315

Ser Leu Thr Asp Phe Val Phe Val Ala Gly Asn Trp Ile Ser Lys
320 325 330

Phe Phe Ser Ser His Thr Arg Gly Ser Leu Asp Pro Tyr Glu Gly
335 340 345

Gln Glu Thr Val Val Arg Ala Met Leu Ala Phe Leu Gln Lys His
350 355 360

Leu Asp Leu Lys Glu Asp Tyr Asp Gln Trp Asn Asn Phe Ile Glu
365 370 375

Gly Ile Gly Pro Ser Leu Thr Pro Gly Ala Pro His His Leu Ser
380 385 390

Ser Leu
392

SEQ. ID. No.: 2

SEQ. LENGTH: 1665

SEQ. TYPE: nucleic acid

MOLECULE TYPE: cDNA

ORIGINAL SOURCE:

ORGANISM: bovine (*Bos taurus*)

SEQUENCE DESCRIPTION:

```
                GTCGACCCACGCGTCCGAGTTGACCGTCTGGGCTGTTTCTGAGGGTCAAC   50

GTGACTCGCCGTCAAGTTCAGCCACTGCCCAAGTCGTCGTTCAGTTCAGTTGGTTATGAG  110

ATG GGG GTC AAC CAG TCT GTG AGC TTC CCA CCC GTC ACG GGA CCC    155
Met Gly Val Asn Gln Ser Val Ser Phe Pro Pro Val Thr Gly Pro
1               5                  10                  15

CAC CTC GTA GGC TGT GGG GAT GTG ATG GAG GGT CAG AGC CTC CAG    200
His Leu Val Gly Cys Gly Asp Val Met Glu Gly Gln Ser Leu Gln
                20                  25                  30

GGC AGC TTC TTT CGA CTG TTC TAC CCG TGC CAA GAG GCA GAG GAG    245
Gly Ser Phe Phe Arg Leu Phe Tyr Pro Cys Gln Glu Ala Glu Glu
                35                  40                  45

ACC TCG GAG CAG CCC CTG TGG ATT CCC CGC TAT GAG TAC TGC GCT    290
Thr Ser Glu Gln Pro Leu Trp Ile Pro Arg Tyr Glu Tyr Cys Ala
                50                  55                  60
```

GGC CTG GCC GAA TAC CTA AAG TTT AAT AAG CGC TGG GGG GGG TTA 335
Gly Leu Ala Glu Tyr Leu Lys Phe Asn Lys Arg Trp Gly Gly Leu
65 70 75

CTG TTC AAC CTG GGT GTG GGA TCT TGT CGC CTG CCT GTT AGC TGG 380
Leu Phe Asn Leu Gly Val Gly Ser Cys Arg Leu Pro Val Ser Trp
80 85 90

AAT GGC CCC TTT AAA ACA AAG GAC TCT GGA TAC CCC TTG ATC ATC 425
Asn Gly Pro Phe Lys Thr Lys Asp Ser Gly Tyr Pro Leu Ile Ile
95 100 105

TTC TCT CAT GGC ATG GGA GCC TTC AGG ACA GTG TAT TCA GCC TTC 470
Phe Ser His Gly Met Gly Ala Phe Arg Thr Val Tyr Ser Ala Phe
110 115 120

TGC ATG GAG CTG GCT TCT CGT GGC TTT GTG GTT GCT GTA CCA GAG 515
Cys Met Glu Leu Ala Ser Arg Gly Phe Val Val Ala Val Pro Glu
125 130 135

CAC AGG GAT GGG TCA GCT GCG GCC ACC TGT TTC TGC AAG CAG ACC 560
His Arg Asp Gly Ser Ala Ala Ala Thr Cys Phe Cys Lys Gln Thr
140 145 150

CCA GAG GAG AAC CAG CCT GAC AAT GAG GCC CTG AAG GAG GAA TGG 605
Pro Glu Glu Asn Gln Pro Asp Asn Glu Ala Leu Lys Glu Glu Trp
155 160 165

```
ATC CCC CAC CGT CAA ATT GAG GAA GGG GAG AAG GAA TTC TAT GTT   650
Ile Pro His Arg Gln Ile Glu Glu Gly Glu Lys Glu Phe Tyr Val
                170                 175                 180

CGG AAC TAC CAG GTG CAT CAG AGG GTG AGC GAG TGT GTG AGG GTG   695
Arg Asn Tyr Gln Val His Gln Arg Val Ser Glu Cys Val Arg Val
                185                 190                 195

TTG AAG ATC CTA CAA GAG GTC ACT GCT GGG CAG GCC GTT CTC AAC   740
Leu Lys Ile Leu Gln Glu Val Thr Ala Gly Gln Ala Val Leu Asn
                200                 205                 210

ATC TTG CCT GGC GGA TTG GAT CTG ATG ACC TTG AAG GGC GGC ATT   785
Ile Leu Pro Gly Gly Leu Asp Leu Met Thr Leu Lys Gly Gly Ile
                215                 220                 225

GAC GTG AGC CGT GTG GCT GTA ATG GGA CAT TCA TTT GGA GGG GCC   830
Asp Val Ser Arg Val Ala Val Met Gly His Ser Phe Gly Gly Ala
                230                 235                 240

ACA GCT ATT CTG GCC TTG GCC AAG GAG ATG CAA TTT AGG TGT GCT   875
Thr Ala Ile Leu Ala Leu Ala Lys Glu Met Gln Phe Arg Cys Ala
                245                 250                 255

GTG GCT TTG GAC GCT TGG ATG TTT CCT CTG GAG CAT GAC TTT TAC   920
Val Ala Leu Asp Ala Trp Met Phe Pro Leu Glu His Asp Phe Tyr
                260                 265                 270
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CCC ACG GCC CGA GGC CCT ATC TTC TTT ATC AAT GCT GAG AAG TTC   965
Pro Thr Ala Arg Gly Pro Ile Phe Phe Ile Asn Ala Glu Lys Phe
                275                 280                 285

CAG ACA GTG GAG ACT GTC AAC TTG ATG AAA AAG ATT TGT GAC CAG  1010
Gln Thr Val Glu Thr Val Asn Leu Met Lys Lys Ile Cys Asp Gln
                290                 295                 300

CAC CAC CAA TCC AGG ATC ATA ACT GTC CTT GGT TCT GTT CAT CGG  1055
His His Gln Ser Arg Ile Ile Thr Val Leu Gly Ser Val His Arg
                305                 310                 315

AGT CTA ACC GAC TTT GTT TTT GTG GCT GGT AAC TGG ATT AGT AAA  1100
Ser Leu Thr Asp Phe Val Phe Val Ala Gly Asn Trp Ile Ser Lys
                320                 325                 330

TTC TTC TCC AGT CAC ACC CGT GGA AGC TTG GAC CCC TAT GAA GGT  1145
Phe Phe Ser Ser His Thr Arg Gly Ser Leu Asp Pro Tyr Glu Gly
                335                 340                 345

CAG GAG ACC GTG GTG CGG GCC ATG TTG GCC TTC CTG CAG AAG CAT  1190
Gln Glu Thr Val Val Arg Ala Met Leu Ala Phe Leu Gln Lys His
                350                 355                 360

CTT GAC CTG AAA GAG GAC TAT GAC CAG TGG AAC AAC TTC ATT GAA  1235
Leu Asp Leu Lys Glu Asp Tyr Asp Gln Trp Asn Asn Phe Ile Glu
                365                 370                 375
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GGC ATT GGC CCA TCA CTG ACC CCA GGG GCC CCA CAC CAT CTG TCC 1280
Gly Ile Gly Pro Ser Leu Thr Pro Gly Ala Pro His His Leu Ser
380 385 390

AGC CTG TAG GCACAACTGGTCATCTTGTGGAAGGTCCCTGAGCTGAGTTCCCGTGT 1336
Ser Leu
392

GGGGCCTGCCCAGGGATACCCTTGGCCTCCTATCAGGAAGTGATTGCCATGACCCTTCTG 1396

TGTTGATTGAGAGGATATAATCACACTGCTGATTGGTAACGGGGTACTTGGATTCTCAGA 1456

CTTGTCGATCTTAAACTCATGTTGGGACTTGGGTTCACTTACTGATGGGCAAACGGGCAT 1516

TCTGAGGACTGAGCCTTAATGGTATGGAGAACAAACAGTGGGATGGGGCTGGGGAAGATC 1576

TAAGCCCTAAGCTGGGCACTATGAGCCCTATAAACCCAACCAGCCAACACCCTCACCTTG 1636

GGCAAGTATGACTTCTGCAGGTCGACTCT 1665

SEQ. ID. No.: 3

SEQ. LENGTH: 392

SEQ. TYPE: amino acid

MOLECULE TYPE: peptide

ORIGINAL SOURCE:

ORGANISM: human

SEQUENCE DESCRIPTION:

Met	Gly	Val	Asn	Gln	Ser	Val	Gly	Phe	Pro	Pro	Val	Thr	Gly	Pro	
1				5				10						15	
His	Leu	Val	Gly	Cys	Gly	Asp	Val	Met	Glu	Gly	Gln	Asn	Leu	Gln	
				20				25						30	
Gly	Ser	Phe	Phe	Arg	Leu	Phe	Tyr	Pro	Cys	Gln	Lys	Ala	Glu	Glu	
				35				40						45	
Thr	Met	Glu	Gln	Pro	Leu	Trp	Ile	Pro	Arg	Tyr	Glu	Tyr	Cys	Thr	
				50				55						60	
Gly	Leu	Ala	Glu	Tyr	Leu	Gln	Phe	Asn	Lys	Arg	Cys	Gly	Gly	Leu	
				65				70						75	
Leu	Phe	Asn	Leu	Ala	Val	Gly	Ser	Cys	Arg	Leu	Pro	Val	Ser	Trp	
				80				85						90	
Asn	Gly	Pro	Phe	Lys	Thr	Lys	Asp	Ser	Gly	Tyr	Pro	Leu	Ile	Ile	
				95				100						105	
Phe	Ser	His	Gly	Leu	Gly	Ala	Phe	Arg	Thr	Leu	Tyr	Ser	Ala	Phe	
				110				115						120	
Cys	Met	Glu	Leu	Ala	Ser	Arg	Gly	Phe	Val	Val	Ala	Val	Pro	Glu	
				125				130						135	
His	Arg	Asp	Arg	Ser	Ala	Ala	Thr	Thr	Tyr	Phe	Cys	Lys	Gln	Ala	
				140				145						150	

Pro Glu Glu Asn Gln Pro Thr Asn Glu Ser Leu Gln Glu Glu Trp
155 160 165

Ile Pro Phe Arg Arg Val Glu Glu Gly Glu Lys Glu Phe His Val
170 175 180

Arg Asn Pro Gln Val His Gln Arg Val Ser Glu Cys Leu Arg Val
185 190 195

Leu Lys Ile Leu Gln Glu Val Thr Ala Gly Gln Thr Val Phe Asn
200 205 210

Ile Leu Pro Gly Gly Leu Asp Leu Met Thr Leu Lys Gly Asn Ile
215 220 225

Asp Met Ser Arg Val Ala Val Met Gly His Ser Phe Gly Gly Ala
230 235 240

Thr Ala Ile Leu Ala Leu Ala Lys Glu Thr Gln Phe Arg Cys Ala
245 250 255

Val Ala Leu Asp Ala Trp Met Phe Pro Leu Glu Arg Asp Phe Tyr
260 265 270

Pro Lys Ala Arg Gly Pro Val Phe Phe Ile Asn Thr Glu Lys Phe
275 280 285

Gln Thr Met Glu Ser Val Asn Leu Met Lys Lys Ile Cys Ala Gln
290 295 300

His Glu Gln Ser Arg Ile Ile Thr Val Leu Gly Ser Val His Arg
305 310 315

Ser Gln Thr Asp Phe Ala Phe Val Thr Gly Asn Leu Ile Gly Lys
320 325 330

Phe Phe Ser Thr Glu Thr Arg Gly Ser Leu Asp Pro Tyr Glu Gly
335 340 345

Gln Glu Val Met Val Arg Ala Met Leu Ala Phe Leu Gln Lys His
350 355 360
Leu Asp Leu Lys Glu Asp Tyr Asn Gln Trp Asn Asn Leu Ile Glu
365 370 375
Gly Ile Gly Pro Ser Leu Thr Pro Gly Ala Pro His His Leu Ser
380 385 390
Ser Leu
392

SEQ. ID. No.: 4

SEQ. LENGTH: 2559

SEQ. TYPE: nucleic acid

MOLECULE TYPE: cDNA

ORIGINAL SOURCE:

ORGANISM: human

SEQUENCE DESCRIPTION:

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                                GCAGGTCTCGACCCACGCGTCCGCGGACGCGTGGG    35

CGAGAAGTGCTTCCAAGCGTCCATTTTGAGCCTTGGAAACTACGACGACCAAAGGGCCAC    95

GGGTTCCTGGGTCGTTTCTCATTTCCGTCGAGTTAAACGTCTGGGGCTGCTTCTGAGGAA   155

TCAGCTTGGCTGGCCAGCAAGTTCAGCTCCGGCAAGTCATTTGATTCACCCGGTGATGAA   215

ATG GGG GTC AAC CAG TCT GTG GGC TTT CCA CCT GTC ACA GGA CCC      260
Met Gly Val Asn Gln Ser Val Gly Phe Pro Pro Val Thr Gly Pro
1               5                  10                  15

CAC CTC GTA GGC TGT GGG GAT GTG ATG GAG GGT CAG AAT CTC CAG      305
His Leu Val Gly Cys Gly Asp Val Met Glu Gly Gln Asn Leu Gln
                20                  25                  30

GGG AGC TTC TTT CGA CTC TTC TAC CCC TGC CAA AAG GCA GAG GAG      350
Gly Ser Phe Phe Arg Leu Phe Tyr Pro Cys Gln Lys Ala Glu Glu
                35                  40                  45
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ACC ATG GAG CAG CCC CTG TGG ATT CCC CGC TAT GAG TAC TGC ACT 395
Thr Met Glu Gln Pro Leu Trp Ile Pro Arg Tyr Glu Tyr Cys Thr
50 55 60

GGC CTG GCC GAG TAC CTG CAG TTT AAT AAG CGC TGC GGG GGC TTG 440
Gly Leu Ala Glu Tyr Leu Gln Phe Asn Lys Arg Cys Gly Gly Leu
65 70 75

CTG TTC AAC CTG GCG GTG GGA TCT TGT CGC CTG CCT GTT AGC TGG 495
Leu Phe Asn Leu Ala Val Gly Ser Cys Arg Leu Pro Val Ser Trp
80 85 90

AAT GGC CCC TTT AAG ACA AAG GAC TCT GGA TAC CCC TTG ATC ATC 540
Asn Gly Pro Phe Lys Thr Lys Asp Ser Gly Tyr Pro Leu Ile Ile
95 100 105

TTC TCC CAT GGC CTA GGA GCC TTC AGG ACT TTG TAT TCA GCC TTC 585
Phe Ser His Gly Leu Gly Ala Phe Arg Thr Leu Tyr Ser Ala Phe
110 115 120

TGC ATG GAG CTG GCC TCA CGT GGC TTT GTG GTT GCT GTG CCA GAG 630
Cys Met Glu Leu Ala Ser Arg Gly Phe Val Val Ala Val Pro Glu
125 130 135

CAC AGG GAC CGG TCA GCG GCA ACC ACC TAT TTC TGC AAG CAG GCC 675
His Arg Asp Arg Ser Ala Ala Thr Thr Tyr Phe Cys Lys Gln Ala
140 145 150

```
CCA GAA GAG AAC CAG CCC ACC AAT GAA TCG CTG CAG GAG GAA TGG   720
Pro Glu Glu Asn Gln Pro Thr Asn Glu Ser Leu Gln Glu Glu Trp
                155                 160                 165

ATC CCT TTC CGT CGA GTT GAG GAA GGG GAG AAG GAA TTT CAT GTT   765
Ile Pro Phe Arg Arg Val Glu Glu Gly Glu Lys Glu Phe His Val
                170                 175                 180

CGG AAT CCC CAG GTG CAT CAG CGG GTA AGC GAG TGT TTA CGG GTG   810
Arg Asn Pro Gln Val His Gln Arg Val Ser Glu Cys Leu Arg Val
                185                 190                 195

TTG AAG ATC CTG CAA GAG GTC ACT GCT GGG CAG ACT GTC TTC AAC   855
Leu Lys Ile Leu Gln Glu Val Thr Ala Gly Gln Thr Val Phe Asn
                200                 205                 210

ATC TTG CCT GGT GGC TTG GAT CTG ATG ACT TTG AAG GGC AAC ATT   900
Ile Leu Pro Gly Gly Leu Asp Leu Met Thr Leu Lys Gly Asn Ile
                215                 220                 225

GAC ATG AGC CGT GTG GCT GTG ATG GGA CAT TCA TTT GGA GGG GCC   945
Asp Met Ser Arg Val Ala Val Met Gly His Ser Phe Gly Gly Ala
                230                 235                 240

ACA GCT ATT CTG GCT TTG GCC AAG GAG ACC CAA TTT CGG TGT GCG   990
Thr Ala Ile Leu Ala Leu Ala Lys Glu Thr Gln Phe Arg Cys Ala
                245                 250                 255
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GTG GCT CTG GAT GCT TGG ATG TTT CCT CTG GAA CGT GAC TTT TAC 1035
Val Ala Leu Asp Ala Trp Met Phe Pro Leu Glu Arg Asp Phe Tyr
260 265 270

CCC AAG GCC CGA GGA CCT GTG TTC TTT ATC AAT ACT GAG AAA TTC 1080
Pro Lys Ala Arg Gly Pro Val Phe Phe Ile Asn Thr Glu Lys Phe
275 280 285

CAG ACA ATG GAG AGT GTC AAT TTG ATG AAG AAG ATA TGT GCC CAG 1125
Gln Thr Met Glu Ser Val Asn Leu Met Lys Lys Ile Cys Ala Gln
290 295 300

CAT GAA CAG TCT AGG ATC ATA ACC GTT CTT GGT TCT GTT CAT CGG 1170
His Glu Gln Ser Arg Ile Ile Thr Val Leu Gly Ser Val His Arg
305 310 315

AGT CAA ACT GAC TTT GCT TTT GTG ACT GGC AAC TTG ATT GGT AAA 1215
Ser Gln Thr Asp Phe Ala Phe Val Thr Gly Asn Leu Ile Gly Lys
320 325 330

TTC TTC TCC ACT GAA ACC CGT GGG AGC CTG GAC CCC TAT GAA GGG 1260
Phe Phe Ser Thr Glu Thr Arg Gly Ser Leu Asp Pro Tyr Glu Gly
335 340 345

CAG GAG GTT ATG GTA CGG GCC ATG TTG GCC TTC CTG CAG AAG CAC 1305
Gln Glu Val Met Val Arg Ala Met Leu Ala Phe Leu Gln Lys His
350 355 360

```
CTC GAC CTG AAA GAA GAC TAT AAT CAA TGG AAC AAC CTT ATT GAA  1350
Leu Asp Leu Lys Glu Asp Tyr Asn Gln Trp Asn Asn Leu Ile Glu
                365                 370                 375

GGC ATT GGA CCG TCG CTC ACC CCA GGG GCC CCC CAC CAT CTG TCC  1395
Gly Ile Gly Pro Ser Leu Thr Pro Gly Ala Pro His His Leu Ser
                380                 385                 390

AGC CTG TAG GCACAACTGGCCATTTGTAAAGTCACTTCAGCCAAGTTTTCATTTGGG 1452
Ser Leu  *
    392

AGCTACCCAAGGGCACCCATGAGCTCCTATCAAGAAGTGATCAACGTGACCCCTTTTCAC 1512

AGATTGAAAGGTGTAATCACACTGCTGCTTGGATAACTGGGTACTTTGATCTTAGATTTG 1572

ATCTTAAAATCACTTTGGGACTGGGATCCCTTGCTGATTGACAAACAGACTTTCTGGGAC 1632

CTTGATGGAGTGGGGAACAAGCAGTAGAGTGGGACTGGGGGAGACCCAGGCCCCGGGCTG 1692

AGCACTGTGAGGCCTGGATGTGAAGACTCAGCCCAGCGAAGCTCATTCCCTTACCCCCGG 1752

CCAGTGCTGCTGCTTCAGTGGAAGAGATGAAGCCAAAGGACAGAATGAAAATCCCTACCT 1812

TCAGAGACTCTAGCCCAGCCCAACACCATCTCTTCCTACCTCTCAGCCTTCTCCCTCCCC 1872
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AGGGCCACTTGTTGAAGTCTGAGCACTTTATGTAAATTTCTAGGTGTGAGCCGTGATCAC 1932

ATTTTCTATTTATTTCCAAGTCTTCTCATTGTATGGAACATAGTACTACTTATACTTACA 1992

GTAGTAAGTTATACTTGTGAGCCCACAGAGTGGCAGACAGCATGGCTCTCACAGCACAGG 2052

GAGAAAAACTGAGGTACACAGAGGTACCTCAGAAGCTCTGGATGTCTTTGGGGGTTTTGC 2112

TAAGTGTATCTTGATAGGAAACAACAAAAGCAGGTTGAGATGGGGAAGATGACAGAACAA 2172

CAGTGTTAAATGGCCATTTGCACAGGCCTTTGCCACAACAGAGAAGTAGTTTGGTCAGCT 2232

AAAACTCAGCTGCAGCCTGGACAGTAGAGCGAGACCCCATCTTAAAAATAAAGAAGGCTG 2292

GGCGTGGTGGCTCATGCCTGTAATCCCAGCACTTTGGGAGGCCAAGGCAGGCAGATCACT 2352

TAAGGCCAGGAGTTCAAGACCACCTGGCCAACATGGTGAAACCCCGTCTCTACTAAAAAT 2412

ACAAAAAAATTAGCCTGGCGTAATGGCAGGCGCCTATAATCCCAGCTACTCAGGAGGCTGA 2472

AGCAGAAGAATCACTTGAACCTAGGAGGCGGAGGTTGCAGTGAGTCAAGATCGCGCCACT 2532

GCACTCCAGCCTGGGTGACAGAGCAAGACTCTGTCTT 2569

SEQUENCE LISTING

(1) GENERAL INFORMATION:

(i) APPLICANT:
(A) NAME: SUNTORY LIMITED
(B) STREET: 1-40, Dojimahama 2-chome, Kita-ku, Osaka-shi,
(C) CITY: OSAKA
(E) COUNTRY: JAPAN
(F) POSTAL CODE (ZIP): 530

(ii) TITLE OF INVENTION: PLATELET ACTIVATING FACTOR ACETYLHYDROLASE, AND GENE THEREOF

(iii) NUMBER OF SEQUENCES: 4

(iv) COMPUTER READABLE FORM:
(A) MEDIUM TYPE: Floppy disk
(B) COMPUTER: IBM PC compatible
(C) OPERATING SYSTEM: PC-DOS/MS-DOS
(D) SOFTWARE: PatentIn Release #1.0, Version #1.30 (EPO)

(2) INFORMATION FOR SEQ ID NO: 1:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 392 amino acids
(B) TYPE: amino acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear

(ii) MOLECULE TYPE: peptide

(vi) ORIGINAL SOURCE:
(A) ORGANISM: human

(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 1:

```
Met Gly Val Asn Gln Ser Val Gly Phe Pro Pro Val Thr Gly Pro His
1               5                  10                  15

Leu Val Gly Cys Gly Asp Val Met Glu Gly Gln Asn Leu Gln Gly Ser
            20                  25                  30

Phe Phe Arg Leu Phe Tyr Pro Cys Gln Lys Ala Glu Glu Thr Met Glu
        35                  40                  45

Gln Pro Leu Trp Ile Pro Arg Tyr Glu Tyr Cys Thr Gly Leu Ala Glu
    50                  55                  60

Tyr Leu Gln Phe Asn Lys Arg Cys Gly Gly Leu Leu Phe Asn Leu Ala
65                  70                  75                  80

Val Gly Ser Cys Arg Leu Pro Val Ser Trp Asn Gly Pro Phe Lys Thr
                85                  90                  95

Lys Asp Ser Gly Tyr Pro Leu Ile Ile Phe Ser His Gly Leu Gly Ala
            100                 105                 110

Phe Arg Thr Leu Tyr Ser Ala Phe Cys Met Glu Leu Ala Ser Arg Gly
        115                 120                 125
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Phe Val Val Ala Val Pro Glu His Arg Asp Arg Ser Ala Ala Thr Thr
    130                 135                 140

Tyr Phe Cys Lys Gln Ala Pro Glu Glu Asn Gln Pro Thr Asn Glu Ser
145                 150                 155                 160

Leu Gln Glu Glu Trp Ile Pro Phe Arg Arg Val Glu Glu Gly Glu Lys
                165                 170                 175

Glu Phe His Val Arg Asn Pro Gln Val His Gln Arg Val Ser Glu Cys
            180                 185                 190

Leu Arg Val Leu Lys Ile Leu Gln Glu Val Thr Ala Gly Gln Thr Val
        195                 200                 205

Phe Asn Ile Leu Pro Gly Gly Leu Asp Leu Met Thr Leu Lys Gly Asn
    210                 215                 220

Ile Asp Met Ser Arg Val Ala Val Met Gly His Ser Phe Gly Gly Ala
225                 230                 235                 240

Thr Ala Ile Leu Ala Leu Ala Lys Glu Thr Gln Phe Arg Cys Ala Val
                245                 250                 255

Ala Leu Asp Ala Trp Met Phe Pro Leu Glu Arg Asp Phe Tyr Pro Lys
            260                 265                 270

Ala Arg Gly Pro Val Phe Phe Ile Asn Thr Glu Lys Phe Gln Thr Met
        275                 280                 285

Glu Ser Val Asn Leu Met Lys Lys Ile Cys Ala Gln His Glu Gln Ser
    290                 295                 300

Arg Ile Ile Thr Val Leu Gly Ser Val His Arg Ser Gln Thr Asp Phe
305                 310                 315                 320

Ala Phe Val Thr Gly Asn Leu Ile Gly Lys Phe Phe Ser Thr Glu Thr
                325                 330                 335

Arg Gly Ser Leu Asp Pro Tyr Glu Gly Gln Glu Val Met Val Arg Ala
            340                 345                 350

Met Leu Ala Phe Leu Gln Lys His Leu Asp Leu Lys Glu Asp Tyr Asn
        355                 360                 365

Gln Trp Asn Asn Leu Ile Glu Gly Ile Gly Pro Ser Leu Thr Pro Gly
    370                 375                 380

Ala Pro His His Leu Ser Ser Leu
385                 390
```

(2) INFORMATION FOR SEQ ID NO: 2:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 2559 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: unknown

(ii) MOLECULE TYPE: cDNA

(vi) ORIGINAL SOURCE:
(A) ORGANISM: human

(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 2:

GCAGGTCTCG ACCCACGCGT CCGCGGACGC GTGGGCGAGA AGTGCTTCCA AGCGTCCATT 60

TTGAGCCTTG GAAACTACGA CGACCAAAGG GCCACGGGTT CCTGGGTCGT TTCTCATTTC 120

CGTCGAGTTA AACGTCTGGG GCTGCTTCTG AGGAATCAGC TTGGCTGGCC AGCAAGTTCA 180

GCTCCGGCAA GTCATTTGAT TCACCCGGTG ATGAAATGGG GGTCAACCAG TCTGTGGGCT 240

TTCCACCTGT CACAGGACCC CACCTCGTAG GCTGTGGGGA TGTGATGGAG GGTCAGAATC 300

TCCAGGGGAG CTTCTTTCGA CTCTTCTACC CCTGCCAAAA GGCAGAGGAG ACCATGGAGC 360

AGCCCCTGTG GATTCCCCGC TATGAGTACT GCACTGGCCT GGCCGAGTAC CTGCAGTTTA 420

ATAAGCGCTG CGGGGGCTTG CTGTTCAACC TGGCGGTGGG ATCTTGTCGC CTGCCTGTTA 480

GCTGGAATGG CCCCTTTAAG ACAAAGGACT CTGGATACCC CTTGATCATC TTCTCCCATG 540

GCCTAGGAGC CTTCAGGACT TTGTATTCAG CCTTCTGCAT GGAGCTGGCC TCACGTGGCT 600

TTGTGGTTGC TGTGCCAGAG CACAGGGACC GGTCAGCGGC AACCACCTAT TTCTGCAAGC 660

AGGCCCCAGA AGAGAACCAG CCCACCAATG AATCGCTGCA GGAGGAATGG ATCCCTTTCC 720

GTCGAGTTGA GGAAGGGGAG AAGGAATTTC ATGTTCGGAA TCCCCAGGTG CATCAGCGGG 780

TAAGCGAGTG TTTACGGGTG TTGAAGATCC TGCAAGAGGT CACTGCTGGG CAGACTGTCT 840

TCAACATCTT GCCTGGTGGC TTGGATCTGA TGACTTTGAA GGGCAACATT GACATGAGCC 900

GTGTGGCTGT GATGGGACAT TCATTTGGAG GGGCCACAGC TATTCTGGCT TTGGCCAAGG 960

AGACCCAATT TCGGTGTGCG GTGGCTCTGG ATGCTTGGAT GTTTCCTCTG GAACGTGACT 1020

TTTACCCCAA GGCCCGAGGA CCTGTGTTCT TTATCAATAC TGAGAAATTC CAGACAATGG 1080

AGAGTGTCAA TTTGATGAAG AAGATATGTG CCCAGCATGA ACAGTCTAGG ATCATAACCG 1140

TTCTTGGTTC TGTTCATCGG AGTCAAACTG ACTTTGCTTT TGTGACTGGC AACTTGATTG 1200

GTAAATTCTT CTCCACTGAA ACCCGTGGGA GCCTGGACCC CTATGAAGGG CAGGAGGTTA 1260

TGGTACGGGC CATGTTGGCC TTCCTGCAGA AGCACCTCGA CCTGAAAGAA GACTATAATC 1320

AATGGAACAA CCTTATTGAA GGCATTGGAC CGTCGCTCAC CCCAGGGGCC CCCCACCATC 1380

TGTCCAGCCT GTAGGCACAA CTGGCCATTT GTAAAGTCAC TTCAGCCAAG TTTTCATTTG 1440

GGAGCTACCC AAGGGCACCC ATGAGCTCCT ATCAAGAAGT GATCAACGTG ACCCCTTTTC 1500

ACAGATTGAA AGGTGTAATC ACACTGCTGC TTGGATAACT GGGTACTTTG ATCTTAGATT 1560

TGATCTTAAA ATCACTTTGG GACTGGGATC CCTTGCTGAT TGACAAACAG ACTTTCTGGG 1620

ACCTTGATGG AGTGGGGAAC AAGCAGTAGA GTGGGACTGG GGGAGACCCA GGCCCCGGGC 1680

TGAGCACTGT GAGGCCTGGA TGTGAAGACT CAGCCCAGCG AAGCTCATTC CCTTACCCCC 1740

GGCCAGTGCT GCTGCTTCAG TGGAAGAGAT GAAGCCAAAG GACAGAATGA AAATCCCTAC 1800

CTTCAGAGAC TCTAGCCCAG CCCAACACCA TCTCTTCCTA CCTCTCAGCC TTCTCCCTCC 1860

CCAGGGCCAC TTGTTGAAGT CTGAGCACTT TATGTAAATT TCTAGGTGTG AGCCGTGATC 1920

ACATTTTCTA TTTATTTCCA AGTCTTCTCA TTGTATGGAA CATAGTACTA CTTATACTTA 1980

CAGTAGTAAG TTATACTTGT GAGCCCACAG AGTGGCAGAC AGCATGGCTC TCACAGCACA 2040

GGGAGAAAAA CTGAGGTACA CAGAGGTACC TCAGAAGCTC TGGATGTCTT TGGGGGTTTT 2100

GCTAAGTGTA TCTTGATAGG AAACAACAAA AGCAGGTTGA GATGGGGAAG ATGACAGAAC 2160

AACAGTGTTA AATGGCCATT TGCACAGGCC TTTGCCACAA CAGAGAAGTA GTTTGGTCAG 2220

CTAAAACTCA GCTGCAGCCT GGACAGTAGA GCGAGACCCC ATCTTAAAAA TAAAGAAGGC 2280

TGGGCGTGGT GGCTCATGCC TGTAATCCCA GCACTTTGGG AGGCCAAGGC AGGCAGATCA 2340

CTTAAGGCCA GGAGTTCAAG ACCACCTGGC CAACATGGTG AAACCCCGTC TCTACTAAAA 2400

ATACAAAAAA TTAGCCTGGC GTAATGGCAG GCGCCTATAA TCCCAGCTAC TCAGGAGGCT 2460

GAAGCAGAAG AATCACTTGA ACCTAGGAGG CGGAGGTTGC AGTGAGTCAA GATCGCGCCA 2520

CTGCACTCCA GCCTGGGTGA CAGAGCAAGA CTCTGTCTT 2559

(2) INFORMATION FOR SEQ ID NO: 3:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 392 amino acids
(B) TYPE: amino acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: unknown

(ii) MOLECULE TYPE: cDNA

(vi) ORIGINAL SOURCE:
(A) ORGANISM: bovine (Bos taurus)

(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 3:

```
Met Gly Val Asn Gln Ser Val Ser Phe Pro Pro Val Thr Gly Pro His
1               5                  10                  15

Leu Val Gly Cys Gly Asp Val Met Glu Gly Gln Ser Leu Gln Gly Ser
            20                  25                  30

Phe Phe Arg Leu Phe Tyr Pro Cys Gln Glu Ala Glu Glu Thr Ser Glu
        35                  40                  45

Gln Pro Leu Trp Ile Pro Arg Tyr Glu Tyr Cys Ala Gly Leu Ala Glu
    50                  55                  60

Tyr Leu Lys Phe Asn Lys Arg Trp Gly Gly Leu Leu Phe Asn Leu Gly
65                  70                  75                  80

Val Gly Ser Cys Arg Leu Pro Val Ser Trp Asn Gly Pro Phe Lys Thr
                85                  90                  95

Lys Asp Ser Gly Tyr Pro Leu Ile Ile Phe Ser His Gly Met Gly Ala
            100                 105                 110

Phe Arg Thr Val Tyr Ser Ala Phe Cys Met Glu Leu Ala Ser Arg Gly
        115                 120                 125

Phe Val Val Ala Val Pro Glu His Arg Asp Gly Ser Ala Ala Ala Thr
    130                 135                 140
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Cys Phe Cys Lys Gln Thr Pro Glu Glu Asn Gln Pro Asp Asn Glu Ala
145                 150                 155                 160

Leu Lys Glu Glu Trp Ile Pro His Arg Gln Ile Glu Glu Gly Glu Lys
                165                 170                 175

Glu Phe Tyr Val Arg Asn Tyr Gln Val His Gln Arg Val Ser Glu Cys
            180                 185                 190

Val Arg Val Leu Lys Ile Leu Gln Glu Val Thr Ala Gly Gln Ala Val
        195                 200                 205

Leu Asn Ile Leu Pro Gly Gly Leu Asp Leu Met Thr Leu Lys Gly Gly
    210                 215                 220

Ile Asp Val Ser Arg Val Ala Val Met Gly His Ser Phe Gly Gly Ala
225                 230                 235                 240

Thr Ala Ile Leu Ala Leu Ala Lys Glu Met Gln Phe Arg Cys Ala Val
                245                 250                 255

Ala Leu Asp Ala Trp Met Phe Pro Leu Glu His Asp Phe Tyr Pro Thr
            260                 265                 270

Ala Arg Gly Pro Ile Phe Phe Ile Asn Ala Glu Lys Phe Gln Thr Val
        275                 280                 285

Glu Thr Val Asn Leu Met Lys Lys Ile Cys Asp Gln His His Gln Ser
    290                 295                 300

Arg Ile Ile Thr Val Leu Gly Ser Val His Arg Ser Leu Thr Asp Phe
305                 310                 315                 320

Val Phe Val Ala Gly Asn Trp Ile Ser Lys Phe Phe Ser Ser His Thr
                325                 330                 335

Arg Gly Ser Leu Asp Pro Tyr Glu Gly Gln Glu Thr Val Val Arg Ala
            340                 345                 350

Met Leu Ala Phe Leu Gln Lys His Leu Asp Leu Lys Glu Asp Tyr Asp
        355                 360                 365

Gln Trp Asn Asn Phe Ile Glu Gly Ile Gly Pro Ser Leu Thr Pro Gly
    370                 375                 380

Ala Pro His His Leu Ser Ser Leu
385                 390
```

(2) INFORMATION FOR SEQ ID NO: 4:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 1665 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: circular

(ii) MOLECULE TYPE: cDNA

(vi) ORIGINAL SOURCE:
(A) ORGANISM: bovine (Bos taurus)

(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 4:

GTCGACCCAC GCGTCCGAGT TGACCGTCTG GGCTGTTTCT GAGGGTCAAC GTGACTCGCC 60

GTCAAGTTCA GCCACTGCCC AAGTCGTCGT TCAGTTCAGT TGGTTATGAG ATGGGGGTCA 120

ACCAGTCTGT GAGCTTCCCA CCCGTCACGG GACCCCACCT CGTAGGCTGT GGGGATGTGA 180

TGGAGGGTCA GAGCCTCCAG GGCAGCTTCT TTCGACTGTT CTACCCGTGC CAAGAGGCAG 240

AGGAGACCTC GGAGCAGCCC CTGTGGATTC CCCGCTATGA GTACTGCGCT GGCCTGGCCG 300

AATACCTAAA GTTTAATAAG CGCTGGGGGG GGTTACTGTT CAACCTGGGT GTGGGATCTT 360

GTCGCCTGCC TGTTAGCTGG AATGGCCCCT TTAAAACAAA GGACTCTGGA TACCCCTTGA 420

TCATCTTCTC TCATGGCATG GGAGCCTTCA GGACAGTGTA TTCAGCCTTC TGCATGGAGC 480

TGGCTTCTCG TGGCTTTGTG GTTGCTGTAC CAGAGCACAG GGATGGGTCA GCTGCGGCCA 540

CCTGTTTCTG CAAGCAGACC CCAGAGGAGA ACCAGCCTGA CAATGAGGCC CTGAAGGAGG 600

AATGGATCCC CCACCGTCAA ATTGAGGAAG GGGAGAAGGA ATTCTATGTT CGGAACTACC 660

AGGTGCATCA GAGGGTGAGC GAGTGTGTGA GGGTGTTGAA GATCCTACAA GAGGTCACTG 720

CTGGGCAGGC CGTTCTCAAC ATCTTGCCTG GCGGATTGGA TCTGATGACC TTGAAGGGCG 780

GCATTGACGT GAGCCGTGTG GCTGTAATGG GACATTCATT TGGAGGGGCC ACAGCTATTC 840

TGGCCTTGGC CAAGGAGATG CAATTTAGGT GTGCTGTGGC TTTGGACGCT TGGATGTTTC 900

CTCTGGAGCA TGACTTTTAC CCCACGGCCC GAGGCCCTAT CTTCTTTATC AATGCTGAGA 960

AGTTCCAGAC AGTGGAGACT GTCAACTTGA TGAAAAAGAT TTGTGACCAG CACCACCAAT 1020

CCAGGATCAT AACTGTCCTT GGTTCTGTTC ATCGGAGTCT AACCGACTTT GTTTTTGTGG 1080

CTGGTAACTG GATTAGTAAA TTCTTCTCCA GTCACACCCG TGGAAGCTTG GACCCCTATG 1140

AAGGTCAGGA GACCGTGGTG CGGGCCATGT TGGCCTTCCT GCAGAAGCAT CTTGACCTGA 1200

AAGAGGACTA TGACCAGTGG AACAACTTCA TTGAAGGCAT TGGCCCATCA CTGACCCCAG 1260

GGGCCCCACA CCATCTGTCC AGCCTGTAGG CACAACTGGT CATCTTGTGG AAGGTCCCTG 1320

AGCTGAGTTC CCGTGTGGGG CCTGCCCAGG GATACCCTTG GCCTCCTATC AGGAAGTGAT 1380

TGCCATGACC CTTCTGTGTT GATTGAGAGG ATATAATCAC ACTGCTGATT GGTAACGGGG 1440

TACTTGGATT CTCAGACTTG TCGATCTTAA ACTCATGTTG GGACTTGGGT TCACTTACTG 1500

ATGGGCAAAC GGGCATTCTG AGGACTGAGC CTTAATGGTA TGGAGAACAA ACAGTGGGAT 1560

GGGGCTGGGG AAGATCTAAG CCCTAAGCTG GGCACTATGA GCCCTATAAA CCCAACCAGC 1620

CAACACCCTC ACCTTGGGCA AGTATGACTT CTGCAGGTCG ACTCT 1665

Claims

1. A protein having activities of a human platelet activating factor acetylhydrolase, and represented by an amino acid sequence represented by the following formula (I) or an amino acid sequence having homology therewith:

Met Gly Val Asn Gln Ser Val Gly Phe Pro Pro Val Thr Gly Pro
His Leu Val Gly Cys Gly Asp Val Met Glu Gly Gln Asn Leu Gln
Gly Ser Phe Phe Arg Leu Phe Tyr Pro Cys Gln Lys Ala Glu Glu
Thr Met Glu Gln Pro Leu Trp Ile Pro Arg Tyr Glu Tyr Cys Thr
Gly Leu Ala Glu Tyr Leu Gln Phe Asn Lys Arg Cys Gly Gly Leu
Leu Phe Asn Leu Ala Val Gly Ser Cys Arg Leu Pro Val Ser Trp
Asn Gly Pro Phe Lys Thr Lys Asp Ser Gly Tyr Pro Leu Ile Ile
Phe Ser His Gly Leu Gly Ala Phe Arg Thr Leu Tyr Ser Ala Phe
Cys Met Glu Leu Ala Ser Arg Gly Phe Val Val Ala Val Pro Glu
His Arg Asp Arg Ser Ala Ala Thr Thr Tyr Phe Cys Lys Gln Ala
Pro Glu Glu Asn Gln Pro Thr Asn Glu Ser Leu Gln Glu Glu Trp
Ile Pro Phe Arg Arg Val Glu Glu Gly Glu Lys Glu Phe His Val
Arg Asn Pro Gln Val His Gln Arg Val Ser Glu Cys Leu Arg Val
Leu Lys Ile Leu Gln Glu Val Thr Ala Gly Gln Thr Val Phe Asn
Ile Leu Pro Gly Gly Leu Asp Leu Met Thr Leu Lys Gly Asn Ile
Asp Met Ser Arg Val Ala Val Met Gly His Ser Phe Gly Gly Ala
Thr Ala Ile Leu Ala Leu Ala Lys Glu Thr Gln Phe Arg Cys Ala
Val Ala Leu Asp Ala Trp Met Phe Pro Leu Glu Arg Asp Phe Tyr
Pro Lys Ala Arg Gly Pro Val Phe Phe Ile Asn Thr Glu Lys Phe
Gln Thr Met Glu Ser Val Asn Leu Met Lys Lys Ile Cys Ala Gln

His Glu Gln Ser Arg Ile Ile Thr Val Leu Gly Ser Val His Arg
Ser Gln Thr Asp Phe Ala Phe Val Thr Gly Asn Leu Ile Gly Lys
Phe Phe Ser Thr Glu Thr Arg Gly Ser Leu Asp Pro Tyr Glu Gly
Gln Glu Val Met Val Arg Ala Met Leu Ala Phe Leu Gln Lys His
Leu Asp Leu Lys Glu Asp Tyr Asn Gln Trp Asn Asn Leu Ile Glu
Gly Ile Gly Pro Ser Leu Thr Pro Gly Ala Pro His His Leu Ser
Ser Leu

(I)

2. A DNA encoding said protein of claim 1.

3. An expression vector having said DNA of claim 2.

4. Recombinant host cells transformed by said expression vector of claim 3.

5. A process for the production of a protein having activities of a human platelet activating factor acetylhydrolase, which comprises culturing said recombinant host cells of claim 4 and collecting said protein from the resulting cultured matter.

6. An antibody against said protein of claim 1.

7. A DNA encoding a protein having activities of a bovine platelet activating factor acetylhydrolase, and represented by an amino acid sequence represented by the following formula (III) or an amino acid sequence having homology therewith:

Met Gly Val Asn Gln Ser Val Ser Phe Pro Pro Val Thr Gly Pro
His Leu Val Gly Cys Gly Asp Val Met Glu Gly Gln Ser Leu Gln
Gly Ser Phe Phe Arg Leu Phe Tyr Pro Cys Gln Glu Ala Glu Glu
Thr Ser Glu Gln Pro Leu Trp Ile Pro Arg Tyr Glu Tyr Cys Ala
Gly Leu Ala Glu Tyr Leu Lys Phe Asn Lys Arg Trp Gly Gly Leu
Leu Phe Asn Leu Gly Val Gly Ser Cys Arg Leu Pro Val Ser Trp
Asn Gly Pro Phe Lys Thr Lys Asp Ser Gly Tyr Pro Leu Ile Ile
Phe Ser His Gly Met Gly Ala Phe Arg Thr Val Tyr Ser Ala Phe
Cys Met Glu Leu Ala Ser Arg Gly Phe Val Val Ala Val Pro Glu
His Arg Asp Gly Ser Ala Ala Ala Thr Cys Phe Cys Lys Gln Thr
Pro Glu Glu Asn Gln Pro Asp Asn Glu Ala Leu Lys Glu Glu Trp
Ile Pro His Arg Gln Ile Glu Glu Gly Glu Lys Glu Phe Tyr Val
Arg Asn Tyr Gln Val His Gln Arg Val Ser Glu Cys Val Arg Val
Leu Lys Ile Leu Gln Glu Val Thr Ala Gly Gln Ala Val Leu Asn
Ile Leu Pro Gly Gly Leu Asp Leu Met Thr Leu Lys Gly Gly Ile
Asp Val Ser Arg Val Ala Val Met Gly His Ser Phe Gly Gly Ala
Thr Ala Ile Leu Ala Leu Ala Lys Glu Met Gln Phe Arg Cys Ala
Val Ala Leu Asp Ala Trp Met Phe Pro Leu Glu His Asp Phe Tyr
Pro Thr Ala Arg Gly Pro Ile Phe Phe Ile Asn Ala Glu Lys Phe
Gln Thr Val Glu Thr Val Asn Leu Met Lys Lys Ile Cys Asp Gln
His His Gln Ser Arg Ile Ile Thr Val Leu Gly Ser Val His Arg
Ser Leu Thr Asp Phe Val Phe Val Ala Gly Asn Trp Ile Ser Lys
Phe Phe Ser Ser His Thr Arg Gly Ser Leu Asp Pro Tyr Glu Gly
Gln Glu Thr Val Val Arg Ala Met Leu Ala Phe Leu Gln Lys His
Leu Asp Leu Lys Glu Asp Tyr Asp Gln Trp Asn Asn Phe Ile Glu
Gly Ile Gly Pro Ser Leu Thr Pro Gly Ala Pro His His Leu Ser
Ser Leu

(III)

8. An expression vector having said DNA of claim 7.

9. Recombinant eucaryotic host cells transformed by said expression vector of claim 8.

10. A process for the production of a protein having activities of a bovine platelet activating factor acetylhydrolase, which comprises culturing said recombinant eucaryotic host cells of claim 9 and collecting said protein from the resulting cultured matter.

11. An antibody against a protein having activities of a bovine platelet activating factor acetylhydrolase, and represented by an amino acid sequence represented by the following formula (III) or an amino acid sequence having homology therewith:

Met Gly Val Asn Gln Ser Val Ser Phe Pro Pro Val Thr Gly Pro
His Leu Val Gly Cys Gly Asp Val Met Glu Gly Gln Ser Leu Gln
Gly Ser Phe Phe Arg Leu Phe Tyr Pro Cys Gln Glu Ala Glu Glu
Thr Ser Glu Gln Pro Leu Trp Ile Pro Arg Tyr Glu Tyr Cys Ala
Gly Leu Ala Glu Tyr Leu Lys Phe Asn Lys Arg Trp Gly Gly Leu
Leu Phe Asn Leu Gly Val Gly Ser Cys Arg Leu Pro Val Ser Trp
Asn Gly Pro Phe Lys Thr Lys Asp Ser Gly Tyr Pro Leu Ile Ile
Phe Ser His Gly Met Gly Ala Phe Arg Thr Val Tyr Ser Ala Phe
Cys Met Glu Leu Ala Ser Arg Gly Phe Val Val Ala Val Pro Glu
His Arg Asp Gly Ser Ala Ala Ala Thr Cys Phe Cys Lys Gln Thr

Pro Glu Glu Asn Gln Pro Asp Asn Glu Ala Leu Lys Glu Glu Trp
Ile Pro His Arg Gln Ile Glu Glu Gly Glu Lys Glu Phe Tyr Val
Arg Asn Tyr Gln Val His Gln Arg Val Ser Glu Cys Val Arg Val
Leu Lys Ile Leu Gln Glu Val Thr Ala Gly Gln Ala Val Leu Asn
Ile Leu Pro Gly Gly Leu Asp Leu Met Thr Leu Lys Gly Gly Ile
Asp Val Ser Arg Val Ala Val Met Gly His Ser Phe Gly Gly Ala
Thr Ala Ile Leu Ala Leu Ala Lys Glu Met Gln Phe Arg Cys Ala
Val Ala Leu Asp Ala Trp Met Phe Pro Leu Glu His Asp Phe Tyr
Pro Thr Ala Arg Gly Pro Ile Phe Phe Ile Asn Ala Glu Lys Phe
Gln Thr Val Glu Thr Val Asn Leu Met Lys Lys Ile Cys Asp Gln
His His Gln Ser Arg Ile Ile Thr Val Leu Gly Ser Val His Arg
Ser Leu Thr Asp Phe Val Phe Val Ala Gly Asn Trp Ile Ser Lys
Phe Phe Ser Ser His Thr Arg Gly Ser Leu Asp Pro Tyr Glu Gly
Gln Glu Thr Val Val Arg Ala Met Leu Ala Phe Leu Gln Lys His
Leu Asp Leu Lys Glu Asp Tyr Asp Gln Trp Asn Asn Phe Ile Glu
Gly Ile Gly Pro Ser Leu Thr Pro Gly Ala Pro His His Leu Ser
Ser Leu

(III)